

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions and listings of claims in the application:

LISTING OF CLAIMS:

1. (currently amended): An enzyme electrode comprising:
an electrode formed on an insulating substrate;
an immobilized enzyme layer formed over the electrode;
an adhesion layer comprising a silane-containing compound formed over the immobilized enzyme layer; and
a permeation-limiting layer comprising a fluorine-containing polymer having a structure where a pendant group containing at least a fluoroalkylene block is attached to an unfluorinated vinyl-based polymer, which is formed on the adhesion layer,
~~wherein the fluorine-containing polymer contains no other fluorine than the fluorines contained in the fluoroalkylene block~~
wherein said fluorine-containing polymer is
(i) a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer thereof; or
(ii) a copolymer composed of a fluoroalcohol ester of a polycarboxylic acid (A) and an alkylalcohol ester of a polycarboxylic acid (B),
in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and

the polycarboxylic acid (A) portion and the polycarboxylic acid (B) portion composes the unfluorinated vinyl-based polymer thereof; and
the adhesion layer is formed only on the immobilized enzyme layer.

2. (original): An enzyme electrode as claimed in Claim 1 wherein said adhesion layer is a layer composed essentially of a silane coupling agent.

3. (previously presented): An enzyme electrode as claimed in Claim 1 or 2 wherein said fluorine-containing polymer is a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer thereof.

4. (previously presented): An enzyme electrode as claimed in Claim 1 or 2 wherein said fluorine-containing polymer is a mixture that contains a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer and additionally an alkylalcohol ester of a polycarboxylic acid (B).

5. (previously presented): An enzyme electrode as claimed in Claim 1 wherein said fluorine-containing polymer is a copolymer composed of a fluoroalcohol ester of a polycarboxylic acid (A) and an alkylalcohol ester of a polycarboxylic acid (B),

in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and

the polycarboxylic acid (A) portion and the polycarboxylic acid (B) portion composes the unfluorinated vinyl-based polymer thereof.

6. (previously presented): An enzyme electrode as claimed in Claim 4 wherein said polycarboxylic acid (B) is selected from polymethacrylic acid, polyacrylic acid or a copolymer of acrylic acid and methacrylic acid.

7. (previously presented): An enzyme electrode as claimed in Claim 3 wherein said polycarboxylic acid (A) is selected from polymethacrylic acid, polyacrylic acid or a copolymer of acrylic acid and methacrylic acid.

8. (withdrawn – currently amended): An enzyme electrode comprising:
an electrode formed on an insulating substrate;
an electrode protective layer covering the electrode;
a binding layer comprising a silane-containing compound, which is formed on the electrode protective layer;

an ion-exchange resin film layer formed on the binding layer;

an immobilized enzyme layer formed on the ion-exchange resin film layer;

an adhesion layer comprising a silane-containing compound, which is formed on the immobilized enzyme layer; and

a permeation-limiting layer comprising a fluorine-containing polymer having a structure where a pendant group containing at least a fluoroalkylene block is attached to an unfluorinated vinyl-based polymer, which is formed on the adhesion layer,

~~wherein the fluorine-containing polymer contains no other fluorine than the fluorines contained in the fluoroalkylene block~~

wherein said fluorine-containing polymer is

(i) a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof; and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer thereof; or

(ii) a copolymer composed of a fluoroalcohol ester of a polycarboxylic acid (A) and an alkylalcohol ester of a polycarboxylic acid (B),

in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof; and

the polycarboxylic acid (A) portion and the polycarboxylic acid (B) portion composes the unfluorinated vinyl-based polymer thereof; and

the adhesion layer is formed only on the immobilized enzyme layer.

9. (withdrawn): An enzyme electrode as claimed in Claim 8 wherein said electrode protective layer is made essentially of a urea compound.

10. (withdrawn): An enzyme electrode as claimed in Claim 8 wherein said binding layer and said adhesion layer are layers composed essentially of a silane coupling agent.

11. (withdrawn): An enzyme electrode as claimed in Claim 8 wherein said fluorine-containing polymer is a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer thereof.

12. (withdrawn): An enzyme electrode as claimed in Claim 8 wherein said fluorine-containing polymer is a mixture that contains the fluoroalcohol ester of the polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer and additionally an alkylalcohol ester of a polycarboxylic acid (B).

13. (withdrawn): An enzyme electrode as claimed in Claim 8, wherein said fluorine-containing polymer is a copolymer composed of a fluoroalcohol ester of a polycarboxylic acid (A) and an alkylalcohol ester of a polycarboxylic acid (B),

in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and

the polycarboxylic acid (A) portion and the polycarboxylic acid (B) portion composes the unfluorinated vinyl-based polymer thereof.

14. (withdrawn): An enzyme electrode as claimed in Claim 12 or 13 wherein said polycarboxylic acid (B) is selected from polymethacrylic acid, polyacrylic acid or a copolymer of acrylic acid and methacrylic acid.

15. (withdrawn): The enzyme electrode as claimed in any one of Claims 11 to 13 wherein said polycarboxylic acid (A) is selected from polymethacrylic acid, polyacrylic acid or a copolymer of acrylic acid and methacrylic acid.

16. (currently amended): An enzyme electrode comprising:
an electrode formed on an insulating substrate;
an immobilized enzyme layer formed on the electrode;
an adhesion layer comprising a silane-containing compound, which is formed on the immobilized enzyme layer; and
a permeation-limiting layer formed on the adhesion layer ;
wherein said permeation-limiting layer consists of a film essentially comprising a fluorine-containing polymer having a structure where a pendant group containing at least a

fluoroalkylene block is attached to an unfluorinated vinyl-based polymer, and an average thickness of said permeation-limiting layer is selected within a range of 0.01 to 1 μm ; and a plurality of grooves with depth selected in a range of 0.1 to 100 nm, which is due to variation of the thickness of said permeation-limiting layer, are present only on the outer surface of said permeation-limiting layer, and

~~the fluorine-containing polymer contains no other fluorine than the fluorines contained in the fluoroalkylene block~~

wherein said fluorine-containing polymer is

(i) a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer thereof; or

(ii) a copolymer composed of a fluoroalcohol ester of a polycarboxylic acid (A) and an alkylalcohol ester of a polycarboxylic acid (B),

in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and

the polycarboxylic acid (A) portion and the polycarboxylic acid (B) portion composes the unfluorinated vinyl-based polymer thereof; and

the adhesion layer is formed only on the immobilized enzyme layer.

17. (previously presented): An enzyme electrode as claimed in Claim 16 wherein an average thickness of said permeation-limiting layer is selected within a range of 0.02 to 0.5 μm ;

and the surface of the permeation-limiting layer has an irregular shape having a surface roughness within a range of 0.0001 to 1, which surface roughness is defined as a ratio of mean value of the variation of the thickness to said average thickness of the permeation-limiting layer.

18. (previously presented): An enzyme electrode as claimed in Claim 16 wherein said adhesion layer is a layer composed essentially of a silane coupling agent.

19. (previously presented): An enzyme electrode as claimed in Claim 16 or 18 wherein said fluorine-containing polymer is a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer thereof.

20. (previously presented): An enzyme electrode as claimed in Claim 16 or 18 wherein said fluorine-containing polymer is a mixture that contains the fluoroalcohol ester of the polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer and additionally an alkylalcohol ester of a polycarboxylic acid (B).

21. (previously presented): An enzyme electrode as claimed in Claim 16 wherein said fluorine-containing polymer is a copolymer composed of a fluoroalcohol ester of a polycarboxylic acid (A) and an alkylalcohol ester of a polycarboxylic acid (B),

in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and

the polycarboxylic acid (A) portion and the polycarboxylic acid (B) portion composes the unfluorinated vinyl-based polymer thereof.

22. (previously presented): An enzyme electrode as claimed in Claim 20 wherein said polycarboxylic acid (B) is selected from polymethacrylic acid, polyacrylic acid or a copolymer of acrylic acid and methacrylic acid.

23. (previously presented): An enzyme electrode as claimed in Claim 19 wherein said polycarboxylic acid (A) is selected from polymethacrylic acid, polyacrylic acid or a copolymer of acrylic acid and methacrylic acid.

24. (withdrawn – currently amended): A process for manufacturing an enzyme electrode comprising the steps of:

forming an electrode film on the main surface of an insulating substrate and then patterning the electrode film to form a plurality of electrodes;

forming an electrode protective layer covering the electrode surface;

forming a binding layer comprising a silane-containing compound on the main surface of the insulating substrate;

forming an ion-exchange resin film layer on the main surface of the insulating substrate;

applying a liquid containing an enzyme to the main surface of the insulating substrate and then drying the insulating substrate to form an immobilized enzyme layer;

applying a liquid comprising a silane-containing compound to the main surface of the insulating substrate and then drying the insulating substrate to form an adhesion layer, and sequentially applying a liquid containing a fluorine-containing polymer to the upper surface of the adhesion layer coating the main surface of the insulating substrate and then drying the insulating substrate to form the permeation-limiting layer; and

dicing the insulating substrate to give a plurality of enzyme electrodes;

wherein said permeation-limiting layer consists of a film comprising a fluorine-containing polymer having a structure where a pendant group containing at least a fluoroalkylene block is attached to an unfluorinated vinyl-based polymer,

an average thickness of said permeation-limiting layer is selected within a range of 0.01 to 1 μm ; and a plurality of grooves with depth selected in a range of 0.1 to 100 nm, which is due to variation of the thickness of said permeation-limiting layer, are present only on the outer surface of the permeation-limiting layer, and

~~the fluorine-containing polymer contains no other fluorine than the fluorines contained in the fluoroalkylene block~~

wherein said fluorine-containing polymer is

(i) a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer thereof; or

(ii) a copolymer composed of a fluoroalcohol ester of a polycarboxylic acid (A) and an alkylalcohol ester of a polycarboxylic acid (B),

in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and

the polycarboxylic acid (A) portion and the polycarboxylic acid (B) portion composes the unfluorinated vinyl-based polymer thereof; and

the adhesion layer is formed only on the immobilized enzyme layer.

25. (withdrawn – currently amended): A process for manufacturing an enzyme electrode comprising the steps of:

forming an electrode film on the main surface of an insulating substrate and then patterning the electrode film to form a plurality of electrodes;

forming an electrode protective layer covering the electrode surface;

forming a binding layer comprising a silane-containing compound on the main surface of the insulating substrate;

forming an ion-exchange resin film layer on the main surface of the insulating substrate;

applying an liquid containing an enzyme to the main surface of the insulating substrate and then drying the insulating substrate to form an immobilized enzyme layer;

applying a liquid comprising a silane-containing compound to the main surface of the insulating substrate and then drying the insulating substrate to form an adhesion layer, and sequentially applying a liquid containing a fluorine-containing polymer to the upper surface of the adhesion layer coating the main surface of the insulating substrate and then drying the insulating substrate to form a permeation-limiting layer; and

licing the insulating substrate to give a plurality of enzyme electrodes,
wherein said permeation-limiting layer consists of a film comprising a fluorine-containing polymer having a structure where a pendant group containing at least a fluoroalkylene block is attached to an unfluorinated vinyl-based polymer, and

~~the fluorine-containing polymer contains no other fluorine than the fluorines contained in the fluoroalkylene block~~

said fluorine-containing polymer is

(i) a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer thereof; or

(ii) a copolymer composed of a fluoroalcohol ester of a polycarboxylic acid (A) and an alkylalcohol ester of a polycarboxylic acid (B),

in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and

the polycarboxylic acid (A) portion and the polycarboxylic acid (B) portion composes the unfluorinated vinyl-based polymer thereof; and

the adhesion layer is formed only on the immobilized enzyme layer.

26. (withdrawn): A process for manufacturing an enzyme electrode as claimed in Claim 25 wherein said permeation-limiting layer is a layer being formed by spin coating.

27. (withdrawn): A process for manufacturing an enzyme electrode as claimed in Claim 25 wherein said silane-containing compound used for forming the adhesion layer is a silane coupling agent.

28. (previously presented): A biosensor comprising an enzyme electrode as claimed in Claim 1 or 16.

29. (canceled).

30. (withdrawn – currently amended): A process for manufacturing an enzyme electrode as claimed in Claim 25 ~~or 29~~ wherein said fluorine-containing polymer is a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer thereof.

31. (withdrawn): A process for manufacturing an enzyme electrode as claimed in Claim 25 wherein said film comprising fluorine-containing polymer is a mixture that contains a fluoroalcohol ester of a polycarboxylic acid (A) in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and the polycarboxylic acid (A) is contained as the unfluorinated vinyl-based polymer and additionally an alkylalcohol ester of a polycarboxylic acid (B).

32. (withdrawn): A process for manufacturing an enzyme electrode as claimed in Claim 25 wherein said fluorine-containing polymer is a copolymer of a fluoroalcohol ester of a polycarboxylic acid (A) and an alkylalcohol ester of a polycarboxylic acid (B),

in which the fluoroalcohol portion is contained as the pendant group containing at least a fluoroalkylene block thereof, and

the polycarboxylic acid (A) portion and the polycarboxylic acid (B) portion composes the unfluorinated vinyl-based polymer thereof.

33. (withdrawn): A process for manufacturing an enzyme electrode as claimed in Claim 31 or 32 wherein said polycarboxylic acid (B) is selected from polymethacrylic acid, polyacrylic acid or a copolymer of acrylic acid and methacrylic acid.

34. (withdrawn): A process for manufacturing an enzyme electrode as claimed in Claim 30 wherein said polycarboxylic acid (A) is selected from polymethacrylic acid, polyacrylic acid or a copolymer of acrylic acid and methacrylic acid.

35. (currently amended): An enzyme electrode as claimed in Claim 1, wherein the adhesion layer comprising the silane-containing compound is formed by using a solution of the silane-containing compound in which a solvent used therein is chosen from the solvent group consisting of pure water and mixtures ~~diluted with pure water~~ of alcohols or esters with pure water in a final concentration for the alcohols or esters of 5 % ~~or less~~.

36. (withdrawn - currently amended): An enzyme electrode as claimed in Claim 8, wherein the adhesion layer comprising the silane-containing compound is formed by using a solution of the silane-containing compound in which a solvent used therein is chosen from the solvent group consisting of pure water and mixtures ~~diluted with pure water~~ of alcohols or esters with pure water in a final concentration for the alcohols or esters of 5 % ~~or less~~.

37. (currently amended): An enzyme electrode as claimed in Claim 16, wherein the adhesion layer comprising the silane-containing compound is formed by using a solution of the silane-containing compound in which a solvent used therein is chosen from the solvent group consisting of pure water and mixtures ~~diluted with pure water~~ of alcohols or esters with pure water in a final concentration for the alcohols or esters of 5 % ~~or less~~.

38. (withdrawn - currently amended): A process for manufacturing an enzyme electrode according to Claim 24,

wherein the liquid comprising the silane-containing compound is a solution of the silane-containing compound in which a solvent used therein is chosen from the solvent group consisting of pure water and mixtures ~~diluted with pure water~~ of alcohols or esters with pure water in a final concentration for the alcohols or esters of 5 % ~~or less~~.

39. (withdrawn - currently amended): A process for manufacturing an enzyme electrode according to Claim 25,

wherein the liquid comprising the silane-containing compound is a solution of the silane-containing compound in which a solvent used therein is chosen from the solvent group consisting of pure water and mixtures ~~diluted with pure water~~ of alcohols or esters with pure water in a final concentration for the alcohols or esters of 5 % ~~or less~~.